

processing diverse chemical compounds. For the production of compounds within industrial bioreactors, it is optimal to have a uniform dispersal of bacterial cells within solution, but mutations in bacterial cells may generate surface properties leading to cell aggregation. During long-term culture in the laboratory, mutants of the bacterium *A. baylyi* ADP1 originated a distinctive phenotype of cell aggregation. Genome sequencing and the analysis of gene knockouts showed this aggregation to be due to mutations in the *per* and *pgi* genes, and a reduction in bioemulsifier production. Qualitative analysis of Atomic Force Microscopy (AFM) visualizations identified altered appearances of cell surfaces correlating with the difference in cell aggregation phenotype. AFM force spectroscopy experiments were then conducted to compare the adhesive and viscoelastic properties of aggregating cells to non-aggregating cells. The most distinctive difference found for force spectroscopy measurements was for a four-fold difference in nN in adhesion that was attributable to *pgi*. Overall, this experiment has resulted in a multilevel approach for the evaluation and detection of a cell aggregation phenotype in mutant strains of *A. baylyi* ADP1.

#### 2026-Pos Board B163

##### Membrane Environment can Enhance the Interaction of Glycan Binding Protein to Cell Surface Glycan Receptors

Lei Shen<sup>1</sup>, Yini Wang<sup>2</sup>, Chia-I Lin<sup>3</sup>, Hung-wen Liu<sup>3</sup>, Athena Guo<sup>2</sup>, Xiaoyang Zhu<sup>1</sup>.

<sup>1</sup>Department of Chemistry, Columbia University, New York, NY, USA,

<sup>2</sup>MicroSurfaces, Inc., Englewood, NJ, USA, <sup>3</sup>College of Pharmacy, University of Texas at Austin, Austin, TX, USA.

The binding of lectins to glycan receptors on the host cell surface is a key step contributing to the virulence and species specificity of most viruses. This is exemplified by the viral protein hemagglutinin (HA) of the influenza A virus, whose binding specificity is modulated by the linkage pattern of terminal sialic acids on glycan receptors of host epithelial cells. Such specificity dictates whether transmission is confined to a particular animal species or jumps between species. Here we show, using H5N1 avian influenza as a model, that the specific binding of recombinant HA to  $\alpha$ 2-3 linked sialic acids can be enhanced dramatically by interaction with the surface of the lipid membrane. This effect can be quantitatively accounted for by a two-stage process in which weak association of HA with the membrane surface precedes more specific and tighter binding to the glycan receptor. The weak protein-membrane interaction discovered here in the model system may play an important secondary role in the infection and pathogenesis of the influenza A virus.

#### 2027-Pos Board B164

##### Impact of Composition upon Ordered Membrane Domain ("Raft") Formation by Lipids from Pathogenic Bacteria

Zhen Huang.

Biochemistry and Cell Biology, Stony Brook University, Stony Brook, NY, USA.

Co-existing ordered (raft) and disordered membrane domains have been identified in the outer membrane of the pathogenic bacterium *Borrelia burgdorferi*, the bacterium which causes Lyme disease. Co-existing ordered and disordered membranes can also be detected into *B. burgdorferi* lipid extracts. However, unlike eukaryotic cells, *B. burgdorferi* lack sphingolipids, which are crucial component of eukaryotic rafts. In order to understand the basis of domain formation in this organism we have isolated the major lipids of *B. burgdorferi* by thin layer chromatography, and have initiated studies of their physical properties when dispersed in aqueous solutions. We have found that mixtures of the predominant lipids found in *B. burgdorferi*, namely, ACGal, a lipid in which a fatty acyl chain and cholesterol are linked to galactose, monogalactosyldiglyceride (MGalD) with phosphatidylcholine (PC) can form ordered domains with thermal stabilities similar to that in whole lipid extracts. However, for individual lipid aqueous dispersions domain formation and/or stability is very different than in whole lipid extracts. Combinations of *B. burgdorferi* lipids are being studied to identify which lipid are necessary and sufficient for the formation of co-existing ordered and disordered domains in this bacterium and related ones.

#### 2028-Pos Board B165

##### Stabilization of Glycosphingolipid Domains by Palmitoyl Ceramide in Unsaturated Phosphatidylcholine Bilayers

Md. Abdullah Al Sazzad, J. Peter Slotte, Max Lönnfors.

Biochemistry, Dept of Bioscience, Åbo Akademi University, Turku, Finland. Ceramides and glycosphingolipids (GSLs) are minor components in most eukaryotic cells. Since ceramides may be generated in lipid raft like domains by enzyme degradation of sphingomyelin (SM), ceramide/GSL interactions may become relevant in cell membranes. To examine their mutual interactions, we have prepared binary and ternary model bilayer systems composed of a

disordered lipid (unsaturated phosphatidylcholine), and different combinations of saturated sphingolipids (palmitoyl SM, palmitoyl ceramide (PCer), and hydroxylated or non-hydroxylated galactosyl or glucosyl palmitoyl-ceramide (PGalCer or PGlcCer)). We have used trans-parinaric acid (tPA) as a probe to detect the ordered domains formed by the sphingolipids in the phosphatidylcholine bilayer. In binary systems, the PCer formed the most thermostable ordered domains, followed by PGalCer, OH-PGalCer, OH-PGlcCer, and PGlcCer. The PSM domains were the least thermostable. Addition of PCer to the GSL or PSM domains increased their thermostability, with the exception of PGalCer, whose thermostability was unaffected by inclusion of PCer. Lifetime analysis of tPA suggested that all sphingolipid ordered domains became even more ordered in the presence of PCer. We conclude that PCer was able to interact with all the examined sphingolipids and increased packing order in the domains.

#### 2029-Pos Board B166

##### Comparison of Line Tension Measurement Techniques in Phase Separated Multi-Component Lipid Monolayers

Juan TigreLazo<sup>1</sup>, Joan C. Kunz<sup>2</sup>, Vision Bagonza<sup>3</sup>, Andrew H. Nguyen<sup>1</sup>, Emil Eldo<sup>2</sup>, Benjamin L. Stottrup<sup>1</sup>.

<sup>1</sup>Physics, Augsburg College, Minneapolis, MN, USA, <sup>2</sup>Chemistry, Augsburg College, Minneapolis, MN, USA, <sup>3</sup>Biology, Augsburg College, Minneapolis, MN, USA.

Langmuir monolayers of multi-component lipid compositions have been used to study the mixing behavior of sterol-phospholipid systems. Using traditional Langmuir pressure-area isotherms and fluorescence microscopy techniques we compare line tension measurements using two methods of image analysis. Line tension between coexisting phases of sterol-rich and sterol-poor domains can be extracted from a Fourier analysis of domain boundary fluctuations (J. Phys. Chem. B, 111:11091-11094). These measurements will be compared to a recently developed non-perturbative technique based on domain size distribution (Proc. Natl. Acad. Sci. 110:13272-1327). Until now these two measurement techniques have not been compared on the same data set. The compositions studied include 30:70 mixtures of cholesterol and DMPC, DLPC, and DCPC. As well as 25:75 mixtures of 25-hydroxycholesterol DMPC systems.

#### 2030-Pos Board B167

##### The Average Area Per Molecule of Cholesterol/PC-Lipid Bilayers: A Review of Experimental Data and a Physically Inspired Model

Jonathan P. Litz, Sarah L. Keller.

Chemistry, University of Washington, Seattle, WA, USA.

We recently documented that beta-cyclodextrin extracts cholesterol at different rates from supported lipid bilayers containing either DMPC, SOPC, or DOPC [Litz & Keller, BJ, 2013, 93A]. Quantitative measurement of the rate of cholesterol depletion relies on accurate knowledge of the average area per molecule within each bilayer, as does calibration of fixed-area molecular dynamic simulations [e.g. Klauda & Nagle, BJ, 2006, 2796]. A challenge is to integrate a plethora of seemingly incompatible experimental results, which yield significantly different average areas per molecule of PC-lipid/cholesterol bilayers. Historically, disagreements between values derived from x-ray and neutron scattering have been attributed to differences in sensitivity between the two techniques, and more recent approaches have analyzed scattering data from both techniques [e.g. Kucerka & Katsaras, BJ, 2008, 2356]. Here I show that the majority of the data from which area measurements are derived is in agreement, and that most disparity in reported values arises from the choice of difficult-to-measure physical parameters. I provide an estimate of the uncertainty of how the area of a PC-lipid bilayer changes as a function of the mole fraction of cholesterol and derive a physically-inspired, two-parameter model to predict the change. I compare the efficacy of my model with that of the currently preferred four-parameter model [Edholm & Nagle, BJ, 2005, 1827]. I then apply my results to quantitatively report rates of cholesterol depletion from two-component lipid bilayers.

#### 2031-Pos Board B168

##### Cholesterol Bilayer Domain in Phospholipid Bilayer Membranes can be Detected by Confocal Microscope

Marija Raguz<sup>1</sup>, Nada Ilic<sup>2</sup>, Suresh Kumar<sup>3</sup>, Mariusz Zereba<sup>4</sup>,

Laxman Mainali<sup>5</sup>, Witold K. Subczynski<sup>5</sup>.

<sup>1</sup>Medical Physics and Biophysics, University of Split, Split, Croatia,

<sup>2</sup>Physics, University of Split, Split, Croatia, <sup>3</sup>Pathology, Medical College of

Wisconsin, Milwaukee, WI, USA, <sup>4</sup>Ophthalmology, Medical College of

Wisconsin, Milwaukee, WI, USA, <sup>5</sup>Biophysics, Medical College of

Wisconsin, Milwaukee, WI, USA.

The unique feature of the eye lens fiber-cell plasma membrane is its extremely high cholesterol content; cholesterol/phospholipid molar ratio can be as high as